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NOV 23 2001  
TECH CENTER 1600/2900  
11/14/2001  
13:38:59

DATE: 11/14/2001  
TIME: 13:38:59

Input Set : A:\2173-0120.st25.txt  
Output Set: N:\CRF3\11142001\I590375.raw

3 <110> APPLICANT: ENDO, Keiji et al.

5 <120> TITLE OF INVENTION: MUTANT ALPHA-AMYLASES

7 <130> FILE REFERENCE: 2173-0120P

9 <140> CURRENT APPLICATION NUMBER: US 09/590,375

10 <141> CURRENT FILING DATE: 2000-06-09

12 <150> PRIOR APPLICATION NUMBER: JP P1999-163569

13 <151> PRIOR FILING DATE: 1999-06-10

15 <160> NUMBER OF SEQ ID NOS: 23

17 <210> SEQ ID NO: 1

18 <211> LENGTH: 480

19 <212> TYPE: PRT

20 <213> ORGANISM: *Bacillus* sp. KSM-K38

22 <400> SEQUENCE: 1

23 Asp Gly Leu Asn Gly Thr Met Met Gln Tyr Tyr Glu Trp His Leu Glu  
24 5 10 15  
25 Asn Asp Gly Gln His Trp Asn Arg Leu His Asp Asp Ala Ala Ala Leu  
26 20 25 30  
27 Ser Asp Ala Gly Ile Thr Ala Ile Trp Ile Pro Pro Ala Tyr Lys Gly  
28 35 40 45  
29 Asn Ser Gln Ala Asp Val Gly Tyr Gly Ala Tyr Asp Leu Tyr Asp Leu  
30 50 55 60  
31 Gly Glu Phe Asn Gln Lys Gly Thr Val Arg Thr Lys Tyr Gly Thr Lys  
32 65 70 75 80  
33 Ala Gln Leu Glu Arg Ala Ile Gly Ser Leu Lys Ser Asn Asp Ile Asn  
34 85 90 95  
35 Val Tyr Gly Asp Val Val Met Asn His Lys Met Gly Ala Asp Phe Thr  
36 100 105 110  
37 Glu Ala Val Gln Ala Val Gln Val Asn Pro Thr Asn Arg Trp Gln Asp  
38 115 120 125  
39 Ile Ser Gly Ala Tyr Thr Ile Asp Ala Trp Thr Gly Phe Asp Phe Ser  
40 130 135 140  
41 Gly Arg Asn Asn Ala Tyr Ser Asp Phe Lys Trp Arg Trp Phe His Phe  
42 145 150 155 160  
43 Asn Gly Val Asp Trp Asp Gln Arg Tyr Gln Glu Asn His Ile Phe Arg  
44 165 170 175  
45 Phe Ala Asn Thr Asn Trp Asn Trp Arg Val Asp Glu Glu Asn Gly Asn  
46 180 185 190  
47 Tyr Asp Tyr Leu Leu Gly Ser Asn Ile Asp Phe Ser His Pro Glu Val  
48 195 200 205  
49 Gln Asp Glu Leu Lys Asp Trp Gly Ser Trp Phe Thr Asp Glu Leu Asp  
50 210 215 220  
51 Leu Asp Gly Tyr Arg Leu Asp Ala Ile Lys His Ile Pro Phe Trp Tyr  
52 225 230 235 240  
53 Thr Ser Asp Trp Val Arg His Gln Arg Asn Glu Ala Asp Gln Asp Leu  
54 245 250 255  
55 Phe Val Val Gly Glu Tyr Trp Lys Asp Asp Val Gly Ala Leu Glu Phe  
56 260 265 270

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57 Tyr Leu Asp Glu Met Asn Trp Glu Met Ser Leu Phe Asp Val Pro Leu
58      275      280      285
59 Asn Tyr Asn Phe Tyr Arg Ala Ser Gln Gln Gly Gly Ser Tyr Asp Met
60      290      295      300
61 Arg Asn Ile Leu Arg Gly Ser Leu Val Glu Ala His Pro Met His Ala
62 305      310      315      320
63 Val Thr Phe Val Asp Asn His Asp Thr Gln Pro Gly Glu Ser Leu Glu
64      325      330      335
65 Ser Trp Val Ala Asp Trp Phe Lys Pro Leu Ala Tyr Ala Thr Ile Leu
66      340      345      350
67 Thr Arg Glu Gly Gly Tyr Pro Asn Val Phe Tyr Gly Asp Tyr Tyr Gly
68      355      360      365
69 Ile Pro Asn Asp Asn Ile Ser Ala Lys Lys Asp Met Ile Asp Glu Leu
70      370      375      380
71 Leu Asp Ala Arg Gln Asn Tyr Ala Tyr Gly Thr Gln His Asp Tyr Phe
72 385      390      395      400
73 Asp His Trp Asp Val Val Gly Trp Thr Arg Glu Gly Ser Ser Ser Arg
74      405      410      415
75 Pro Asn Ser Gly Leu Ala Thr Ile Met Ser Asn Gly Pro Gly Gly Ser
76      420      425      430
77 Lys Trp Met Tyr Val Gly Arg Gln Asn Ala Gly Gln Thr Trp Thr Asp
78      435      440      445
79 Leu Thr Gly Asn Asn Gly Ala Ser Val Thr Ile Asn Gly Asp Gly Trp
80      450      455      460
81 Gly Glu Phe Phe Thr Asn Gly Gly Ser Val Ser Val Tyr Val Asn Gln
82 465      470      475      480
85 <210> SEQ ID NO: 2
86 <211> LENGTH: 485
87 <212> TYPE: PRT
88 <213> ORGANISM: Bacillus sp. KSM-AP1378
90 <400> SEQUENCE: 2
91 His His Asn Gly Thr Asn Gly Thr Met Met Gln Tyr Phe Glu Trp His
92      5      10      15
93 Leu Pro Asn Asp Gly Asn His Trp Asn Arg Leu Arg Asp Asp Ala Ala
94      20      25      30
95 Asn Leu Lys Ser Lys Gly Ile Thr Ala Val Trp Ile Pro Pro Ala Trp
96      35      40      45
97 Lys Gly Thr Ser Gln Asn Asp Val Gly Tyr Gly Ala Tyr Asp Leu Tyr
98      50      55      60
99 Asp Leu Gly Glu Phe Asn Gln Lys Gly Thr Val Arg Thr Lys Tyr Gly
100 65      70      75      80
101 Thr Arg Ser Gln Leu Gln Gly Ala Val Thr Ser Leu Lys Asn Asn Gly
102      85      90      95
103 Ile Gln Val Tyr Gly Asp Val Val Met Asn His Lys Gly Gly Ala Asp
104      100      105      110
105 Gly Thr Glu Met Val Asn Ala Val Glu Val Asn Arg Ser Asn Arg Asn
106      115      120      125
107 Gln Glu Ile Ser Gly Glu Tyr Thr Ile Glu Ala Trp Thr Lys Phe Asp
108      130      135      140

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```

109 Phe Pro Gly Arg Gly Asn Thr His Ser Asn Phe Lys Trp Arg Trp Tyr
110 145          150          155          160
111 His Phe Asp Gly Thr Asp Trp Asp Gln Ser Arg Gln Leu Gln Asn Lys
112          165          170          175
113 Ile Tyr Lys Phe Arg Gly Thr Gly Lys Ala Trp Asp Trp Glu Val Asp
114          180          185          190
115 Ile Glu Asn Gly Asn Tyr Asp Tyr Leu Met Tyr Ala Asp Ile Asp Met
116          195          200          205
117 Asp His Pro Glu Val Ile Asn Glu Leu Arg Asn Trp Gly Val Trp Tyr
118          210          215          220
119 Thr Asn Thr Leu Asn Leu Asp Gly Phe Arg Ile Asp Ala Val Lys His
120 225          230          235          240
121 Ile Lys Tyr Ser Tyr Thr Arg Asp Trp Leu Thr His Val Arg Asn Thr
122          245          250          255
123 Thr Gly Lys Pro Met Phe Ala Val Ala Glu Phe Trp Lys Asn Asp Leu
124          260          265          270
125 Ala Ala Ile Glu Asn Tyr Leu Asn Lys Thr Ser Trp Asn His Ser Val
126          275          280          285
127 Phe Asp Val Pro Leu His Tyr Asn Leu Tyr Asn Ala Ser Asn Ser Gly
128          290          295          300
129 Gly Tyr Phe Asp Met Arg Asn Ile Leu Asn Gly Ser Val Val Gln Lys
130 305          310          315          320
131 His Pro Ile His Ala Val Thr Phe Val Asp Asn His Asp Ser Gln Pro
132          325          330          335
133 Gly Glu Ala Leu Glu Ser Phe Val Gln Ser Trp Phe Lys Pro Leu Ala
134          340          345          350
135 Tyr Ala Leu Ile Leu Thr Arg Glu Gln Gly Tyr Pro Ser Val Phe Tyr
136          355          360          365
137 Gly Asp Tyr Tyr Gly Ile Pro Thr His Gly Val Pro Ser Met Lys Ser
138          370          375          380
139 Lys Ile Asp Pro Leu Leu Gln Ala Arg Gln Thr Tyr Ala Tyr Gly Thr
140 385          390          395          400
141 Gln His Asp Tyr Phe Asp His His Asp Ile Ile Gly Trp Thr Arg Glu
142          405          410          415
143 Gly Asp Ser Ser His Pro Asn Ser Gly Leu Ala Thr Ile Met Ser Asp
144          420          425          430
145 Gly Pro Gly Gly Asn Lys Trp Met Tyr Val Gly Lys His Lys Ala Gly
146          435          440          445
147 Gln Val Trp Arg Asp Ile Thr Gly Asn Arg Ser Gly Thr Val Thr Ile
148          450          455          460
149 Asn Ala Asp Gly Trp Gly Asn Phe Thr Val Asn Gly Gly Ala Val Ser
150 465          470          475          480
151 Val Trp Val Lys Gln
152          485
155 <210> SEQ ID NO: 3
156 <211> LENGTH: 1753
157 <212> TYPE: DNA
158 <213> ORGANISM: Bacillus sp. KSM-K38
160 <220> FEATURE:

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161 <221> NAME/KEY: sig_peptide
162 <222> LOCATION: (162)..(224)
164 <220> FEATURE:
165 <221> NAME/KEY: mat_peptide
166 <222> LOCATION: (225)..()
168 <220> FEATURE:
169 <221> NAME/KEY: CDS
170 <222> LOCATION: (162)..(1664)
173 <400> SEQUENCE: 3
174 gtatgcgaaa cgatgcgcaa aactgcgcaa ctactagcac tcttcaggga ctaaaccacc      60
176 ttttttccaa aaatgacatc atataaacia atttgtctac caatcactat ttaaagctgt      120
178 ttatgatata tgtaagcggt atcattaaaa ggaggtatatt g atg aga aga tgg gta      176
179                                     Met Arg Arg Trp Val
180                                     -20
182 gta gca atg ttg gca gtg tta ttt tta ttt cct tcg gta gta gtt gca      224
183 Val Ala Met Leu Ala Val Leu Phe Leu Phe Pro Ser Val Val Val Ala
184 -15 -10 -5 -1
186 gat gga ttg aac ggt acg atg atg cag tat tat gag tgg cat ttg gaa      272
187 Asp Gly Leu Asn Gly Thr Met Met Gln Tyr Tyr Glu Trp His Leu Glu
188 1 5 10 15
190 aac gac ggg cag cat tgg aat cgg ttg cac gat gat gcc gca gct ttg      320
191 Asn Asp Gly Gln His Trp Asn Arg Leu His Asp Asp Ala Ala Ala Leu
192 20 25 30
194 agt gat gct ggt att aca gct att tgg att ccg cca gcc tac aaa ggt      368
195 Ser Asp Ala Gly Ile Thr Ala Ile Trp Ile Pro Pro Ala Tyr Lys Gly
196 35 40 45
198 aat agt cag gcg gat gtt ggg tac ggt gca tac gat ctt tat gat tta      416
199 Asn Ser Gln Ala Asp Val Gly Tyr Gly Ala Tyr Asp Leu Tyr Asp Leu
200 50 55 60
202 gga gag ttc aat caa aag ggt act gtt cga acg aaa tac gga act aag      464
203 Gly Glu Phe Asn Gln Lys Gly Thr Val Arg Thr Lys Tyr Gly Thr Lys
204 65 70 75 80
206 gca cag ctt gaa cga gct att ggg tcc ctt aaa tct aat gat atc aat      512
207 Ala Gln Leu Glu Arg Ala Ile Gly Ser Leu Lys Ser Asn Asp Ile Asn
208 85 90 95
210 gta tac gga gat gtc gtg atg aat cat aaa atg gga gct gat ttt acg      560
211 Val Tyr Gly Asp Val Val Met Asn His Lys Met Gly Ala Asp Phe Thr
212 100 105 110
214 gag gca gtg caa gct gtt caa gta aat cca acg aat cgt tgg cag gat      608
215 Glu Ala Val Gln Ala Val Gln Val Asn Pro Thr Asn Arg Trp Gln Asp
216 115 120 125
218 att tca ggt gcc tac acg att gat gcg tgg acg ggt ttc gac ttt tca      656
219 Ile Ser Gly Ala Tyr Thr Ile Asp Ala Trp Thr Gly Phe Asp Phe Ser
220 130 135 140
222 ggg cgt aac aac gcc tat tca gat ttt aag tgg aga tgg ttc cat ttt      704
223 Gly Arg Asn Asn Ala Tyr Ser Asp Phe Lys Trp Arg Trp Phe His Phe
224 145 150 155 160
226 aat ggt gtt gac tgg gat cag cgc tat caa gaa aat cat att ttc cgc      752
227 Asn Gly Val Asp Trp Asp Gln Arg Tyr Gln Glu Asn His Ile Phe Arg

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228	165																170				175				
230	ttt	gca	aat	acg	aac	tgg	aac	tgg	cga	gtg	gat	gaa	gag	aac	ggt	aat	800								
231	Phe	Ala	Asn	Thr	Asn	Trp	Asn	Trp	Arg	Val	Asp	Glu	Glu	Asn	Gly	Asn									
232	180								185				190												
234	tat	gat	tac	ctg	tta	gga	tcg	aat	atc	gac	ttt	agt	cat	cca	gaa	gta	848								
235	Tyr	Asp	Tyr	Leu	Leu	Gly	Ser	Asn	Ile	Asp	Phe	Ser	His	Pro	Glu	Val									
236	195								200				205												
238	caa	gat	gag	ttg	aag	gat	tgg	ggt	agc	tgg	ttt	acc	gat	gag	tta	gat	896								
239	Gln	Asp	Glu	Leu	Lys	Asp	Trp	Gly	Ser	Trp	Phe	Thr	Asp	Glu	Leu	Asp									
240	210								215				220												
242	ttg	gat	ggt	tat	cgt	tta	gat	gct	att	aaa	cat	att	cca	ttc	tgg	tat	944								
243	Leu	Asp	Gly	Tyr	Arg	Leu	Asp	Ala	Ile	Lys	His	Ile	Pro	Phe	Trp	Tyr									
244	225					230				235				240											
246	aca	tct	gat	tgg	gtt	cgg	cat	cag	cgc	aac	gaa	gca	gat	caa	gat	tta	992								
247	Thr	Ser	Asp	Trp	Val	Arg	His	Gln	Arg	Asn	Glu	Ala	Asp	Gln	Asp	Leu									
248	245								250				255												
250	ttt	gtc	gta	ggg	gaa	tat	tgg	aag	gat	gac	gta	ggt	gct	ctc	gaa	ttt	1040								
251	Phe	Val	Val	Gly	Glu	Tyr	Trp	Lys	Asp	Asp	Val	Gly	Ala	Leu	Glu	Phe									
252	260								265				270												
254	tat	tta	gat	gaa	atg	aat	tgg	gag	atg	tct	cta	ttc	gat	gtt	cca	ctt	1088								
255	Tyr	Leu	Asp	Glu	Met	Asn	Trp	Glu	Met	Ser	Leu	Phe	Asp	Val	Pro	Leu									
256	275								280				285												
258	aat	tat	aat	ttt	tac	cgg	gct	tca	caa	caa	ggt	gga	agc	tat	gat	atg	1136								
259	Asn	Tyr	Asn	Phe	Tyr	Arg	Ala	Ser	Gln	Gln	Gly	Gly	Ser	Tyr	Asp	Met									
260	290								295				300												
262	cgt	aat	att	tta	cga	gga	tct	tta	gta	gaa	gcg	cat	ccg	atg	cat	gca	1184								
263	Arg	Asn	Ile	Leu	Arg	Gly	Ser	Leu	Val	Glu	Ala	His	Pro	Met	His	Ala									
264	305					310				315				320											
266	gtt	acg	ttt	gtt	gat	aat	cat	gat	act	cag	cca	ggg	gag	tca	tta	gag	1232								
267	Val	Thr	Phe	Val	Asp	Asn	His	Asp	Thr	Gln	Pro	Gly	Glu	Ser	Leu	Glu									
268	325								330				335												
270	tca	tgg	gtt	gct	gat	tgg	ttt	aag	cca	ctt	gct	tat	gcg	aca	att	ttg	1280								
271	Ser	Trp	Val	Ala	Asp	Trp	Phe	Lys	Pro	Leu	Ala	Tyr	Ala	Thr	Ile	Leu									
272	340								345				350												
274	acg	cgt	gaa	ggt	ggt	tat	cca	aat	gta	ttt	tac	ggt	gat	tac	tat	ggg	1328								
275	Thr	Arg	Glu	Gly	Gly	Tyr	Pro	Asn	Val	Phe	Tyr	Gly	Asp	Tyr	Tyr	Gly									
276	355								360				365												
278	att	cct	aac	gat	aac	att	tca	gct	aaa	aaa	gat	atg	att	gat	gag	ctg	1376								
279	Ile	Pro	Asn	Asp	Asn	Ile	Ser	Ala	Lys	Lys	Asp	Met	Ile	Asp	Glu	Leu									
280	370								375				380												
282	ctt	gat	gca	cgt	caa	aat	tac	gca	tat	ggc	acg	cag	cat	gac	tat	ttt	1424								
283	Leu	Asp	Ala	Arg	Gln</																				

VERIFICATION SUMMARY

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